

We Claim:

- 000750 520E0460
00403075 054000
1. An isolated nucleic acid molecule selected from the group consisting of:
- a) a nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO:3 or a complement thereof;
 - b) a nucleic acid molecule comprising a fragment of at least 100 contiguous nucleotides of a nucleic acid comprising the nucleotide sequence of SEQ ID NO:3 or a complement thereof;
 - c) a nucleic acid molecule which encodes a polypeptide comprising the amino acid sequence of SEQ ID NO:4;
 - d) a nucleic acid molecule which encodes a fragment of a polypeptide comprising the amino acid sequence of SEQ ID NO:4, wherein the fragment comprises at least 15 contiguous amino acid residues of the amino acid sequence of SEQ ID NO:4;
 - e) a nucleic acid molecule which encodes a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:4, wherein the nucleic acid molecule hybridizes to a nucleic acid molecule comprising SEQ ID NO:3 under stringent conditions; and
 - f) a nucleic acid molecule which is antisense to the coding strand of a nucleic acid molecule having the nucleotide sequence of SEQ ID NO:3.
2. The nucleic acid molecule of claim 1 further comprising vector nucleic acid sequences.
3. The nucleic acid molecule of claim 1 further comprising nucleic acid sequences encoding a heterologous polypeptide.
4. A host cell which contains the nucleic acid molecule of claim 1.
-
5. An isolated nucleic acid molecule selected from the group consisting of:
- a) a nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO:5 or a complement thereof;
 - b) a nucleic acid molecule comprising a fragment of at least 100 contiguous nucleotides of a nucleic acid comprising the nucleotide sequence of SEQ ID NO:5 or a complement thereof;
 - c) a nucleic acid molecule which encodes a polypeptide comprising the amino acid sequence of SEQ ID NO:6;

d) a nucleic acid molecule which encodes a fragment of a polypeptide comprising the amino acid sequence of SEQ ID NO:6, wherein the fragment comprises at least 15 contiguous amino acid residues of the amino acid sequence of SEQ ID NO:6;

e) a nucleic acid molecule which encodes a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:6, wherein the nucleic acid molecule hybridizes to a nucleic acid molecule comprising SEQ ID NO:5 under stringent conditions; and

f) a nucleic acid molecule which is antisense to the coding strand of a nucleic acid molecule having the nucleotide sequence of SEQ ID NO:5.

6. The nucleic acid molecule of claim 5 further comprising vector nucleic acid sequences.

7. The nucleic acid molecule of claim 5 further comprising nucleic acid sequences encoding a heterologous polypeptide.

8. A host cell which contains the nucleic acid molecule of claim 5.

9. An isolated polypeptide selected from the group consisting of:

a) a fragment of a polypeptide comprising the amino acid sequence of SEQ ID NO:4, wherein the fragment comprises at least 15 contiguous amino acids of SEQ ID NO:4;

b) a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:4, wherein the polypeptide is encoded by a nucleic acid molecule which hybridizes to a nucleic acid molecule comprising SEQ ID NO:3 under stringent conditions;

c) a polypeptide which is encoded by the nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO:3; and

d) a polypeptide comprising the amino acid sequence of SEQ ID NO:4.

10. The fusion protein comprising the polypeptide of claim 9 operatively
35 linked to heterologous amino acid sequences.

11. /An antibody which selectively binds to a polypeptide of claim 9.

12. A method for producing a polypeptide selected from the group consisting of:

- 5
- a) a polypeptide comprising the amino acid sequence of SEQ ID NO:4;
- b) a fragment of a polypeptide comprising the amino acid sequence of SEQ ID NO:4, wherein the fragment comprises at least 15 contiguous amino acids of SEQ ID NO:4; and
- 10
- c) a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:4, wherein the polypeptide is encoded by a nucleic acid molecule which hybridizes to a nucleic acid molecule comprising SEQ ID NO:3 under stringent conditions;

15 comprising culturing the host cell of claim 4 under conditions in which the nucleic acid molecule is expressed.

13. An isolated polypeptide selected from the group consisting of:

- 20
- a) a fragment of a polypeptide comprising the amino acid sequence of SEQ ID NO:6, wherein the fragment comprises at least 15 contiguous amino acids of SEQ ID NO:6;
- b) a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:6, wherein the polypeptide is encoded by a nucleic acid molecule which hybridizes to a nucleic acid molecule comprising SEQ ID NO:5 under stringent conditions;
- 25
- c) a polypeptide which is encoded by the nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO:5; and
- d) a polypeptide comprising the amino acid sequence of SEQ ID NO:6.

30

14. The fusion protein comprising the polypeptide of claim 13 operatively linked to heterologous amino acid sequences.

15. An antibody which selectively binds to a polypeptide of claim 13.

1103075-054000

16. A method for producing a polypeptide selected from the group consisting of:

- 5
- a) a polypeptide comprising the amino acid sequence of SEQ ID NO:6;
 - b) a fragment of a polypeptide comprising the amino acid sequence of SEQ ID NO:6, wherein the fragment comprises at least 15 contiguous amino acids of SEQ ID NO:6; and
 - c) a naturally occurring allelic variant of a polypeptide comprising the amino acid sequence of SEQ ID NO:6, wherein the polypeptide is encoded by a nucleic acid molecule which hybridizes to a nucleic acid molecule comprising SEQ ID NO:5 under stringent conditions;
- 10

comprising culturing the host cell of claim 8 under conditions in which the nucleic acid molecule is expressed.

15

17. A method for detecting the presence of a MEKK1 polypeptide in a sample comprising:

- 20
- a) contacting the sample with a compound which selectively binds to the polypeptide; and
 - b) determining whether the compound binds to the polypeptide in the sample to thereby detect the presence of a MEKK1 polypeptide in the sample.

18. A kit comprising a compound which selectively binds to a MEKK1 polypeptide and instructions for use.

25

19. A method for detecting the presence of a MEKK1 nucleic acid molecule in a sample comprising:

- 30
- a) contacting the sample with a nucleic acid probe or primer which selectively hybridizes to the nucleic acid molecule; and
 - b) determining whether the nucleic acid probe or primer binds to a nucleic acid molecule in the sample to thereby detect the presence of a MEKK1 nucleic acid molecule in the sample.

35

20. A kit comprising a compound which selectively hybridizes to a MEKK1 nucleic acid molecule and instructions for use.

Ans
B 3

21. A method for detecting the presence of a biological activity of a MEKK1 polypeptide in a sample comprising:

a) contacting the sample with an agent capable of detecting MEKK1 activity; and

5 b) determining the presence of MEKK1 activity in the sample.

22. A method for modulating MEKK1 activity comprising contacting a cell with an agent that modulates MEKK1 activity such that MEKK1 activity in the cell is modulated.

10

23. The method of claim 22 wherein the agent is selected from the group consisting of an antibody that specifically binds to the MEKK1 protein and a nucleic acid molecule having a nucleotide sequence which is antisense to the coding strand of a MEKK1 mRNA of MEKK1 gene.

15

24. A method to treat a subject having a disorder characterized by aberrant MEKK1 protein or nucleic acid expression or activity comprising administering an agent which is a MEKK1 modulator to the subject such that MEKK1 protein or nucleic acid expression or activity is modulated.

20

25. A method for identifying the presence or absence of a genetic alteration characterized by at least one of (i) aberrant modification or mutation of a gene encoding a MEKK1 protein; (ii) mis-regulation of said gene; and (iii) aberrant post-translational modification of a MEKK1 protein, wherein a wild-type form of said gene encodes an protein with a MEKK1 activity

25

26. An isolated active fragment of an MEKK1 protein consisting of an amino acid sequence having at least 75% homology to an amino acid sequence consisting of about amino acids 875-1493 of SEQ ID NO:4, wherein said active fragment mediates apoptosis.

30

27. The active fragment of claim 26, which consists of an amino acid sequence having at least 85% homology to an amino acid sequence consisting of about amino acids 875-1493 of SEQ ID NO:4.

35

000750 92060760

28. The active fragment of claim 26, which consists of an amino acid sequence having at least 95% homology to an amino acid sequence consisting of about amino acids 875-1493 of Figure 9.

5 29. The active fragment of claim 26, which is a mouse MEKK1 active fragment.

30. The active fragment of claim 26, which is a human MEKK1 active fragment.

10 31. The active fragment of claim 26, which is a rat MEKK1 active fragment.

32. The active fragment of claim 26, which consists of about amino acids 875-1493 of SEQ ID NO:4.

15 33. The active fragment of claim 26, which consists of about amino acids 685-1303 of SEQ ID NO:6.

20 34. An isolated protease-resistant MEKK1 protein comprising an amino acid sequence having at least 75% homology to the amino acid sequence of SEQ ID NO:4, wherein at least one amino acid equivalent to amino acids 871-874 of SEQ ID NO:4 is substituted such that the MEKK1 protein is resistant to proteolysis by a caspase after amino acid 874.

25 35. The MEKK1 protein of claim 34, wherein at least one amino acid equivalent to amino acids 871-874 of SEQ ID NO:4 is substituted with an alanine residue.

30 36. The MEKK1 protein of claim 34, wherein each amino acid equivalent to amino acids 871-874 of SEQ ID NO:4 is substituted with an alanine residue.

37. The MEKK1 protein of claim 34, which has at least 85% homology to the amino acid sequence of SEQ ID NO:4.

35 38. The MEKK1 protein of claim 34, which has at least 95% homology to the amino acid sequence of SEQ ID NO:4.

39. The MEKK1 protein of claim 34, which is a mouse MEKK1 protein.
40. The MEKK1 protein of claim 34, which is a human MEKK1 protein.
- 5 41. The MEKK protein of claim 40, consisting of amino acids 685-1303 of SEQ ID NO:6.
42. The MEKK1 protein of claim 34, which is a rat MEKK1 protein.
-
- 10 43. An isolated nucleic acid molecule consisting of a nucleotide sequence having at least 75% homology to a nucleotide sequence consisting of about nucleotides 2637-4493 of SEQ ID NO:3, wherein said nucleic acid molecule encodes an active fragment of MEKK1 that mediates apoptosis.
-
- 15 44. The nucleic acid molecule of claim 43, which consists of a nucleotide sequence having at least 85% homology to a nucleotide sequence consisting of about nucleotides 2637-4493 of SEQ ID NO:3.
45. The nucleic acid molecule of claim 43, which consists of a nucleotide
20 sequence having at least 95% homology to a nucleotide sequence consisting of about nucleotides 2637-4493 of SEQ ID NO:3.
46. The nucleic acid molecule of claim 43, which encodes an active fragment of a mouse MEKK1.
- 25 ^{P.126} 47. The nucleic acid molecule of claim 43, which encodes an active fragment of a human MEKK1.
48. The nucleic acid molecule of claim 43, which encodes an active fragment
30 of a rat MEKK1.
49. The nucleic acid molecule of claim 43, which consists of about nucleotides 2637-4493 of SEQ ID NO:3, or a nucleotide sequence that, due to the degeneracy of the genetic code, encodes the same amino acid sequence as about
35 nucleotides 2637-4493 of SEQ ID NO:3.

000150-92020160

50. The nucleic acid molecule of claim 43, which consists of nucleotides 2637-4493 of SEQ ID NO:3, or a nucleotide sequence that, due to the degeneracy of the genetic code, encodes the same amino acid sequence as nucleotides 2637-4493 of SEQ ID NO:3.

51. The nucleic acid molecule of claim 43, which consists of nucleotides 2052-3908 of SEQ ID NO:5, or a nucleotide sequence that, due to the degeneracy of the genetic code, encodes the same amino acid sequence as nucleotides 2052-3908 of SEQ ID NO:5.

52. An isolated nucleic acid molecule encoding a protease-resistant MEKK1 protein, wherein the protease resistant MEKK1 protein comprises an amino acid sequence having at least 75% homology to the amino acid sequence of SEQ ID NO:4 and at least one codon of the nucleic acid molecule encoding an amino acid equivalent to at least one of amino acids 871-874 of SEQ ID NO:4 is mutated such the encoded MEKK1 protein is resistant to proteolysis by a caspase after an amino acid equivalent to amino acid 871-874 of SEQ ID NO:4.

53. The nucleic acid molecule of claim 52, wherein the MEKK1 protein comprises an amino acid sequence having at least 85% homology to the amino acid sequence of SEQ ID NO:4.

54. The nucleic acid molecule of claim 52, wherein the MEKK1 protein comprises an amino acid sequence having at least 95% homology to the amino acid sequence of SEQ ID NO:4.

55. The nucleic acid molecule of claim 52, which encodes a protease-resistant mouse MEKK1 protein.

56. The nucleic acid molecule of claim 52, which encodes a protease-resistant human MEKK1 protein.

57. The nucleic acid molecule of claim 52, which encodes a protease-resistant rat MEKK1 protein.

58. An expression vector comprising the nucleic acid molecule of claim 43.

Ind
B6
C1

59. An expression vector comprising the nucleic acid molecule of claim 52.

60. A host cell containing the expression vector of claim 58.

61. A host cell containing the expression vector of claim 59.

62. An isolated nucleic acid molecule encoding a protease-resistant MEKK1 protein, wherein the protease-resistant MEKK1 protein comprises the amino acid sequence of SEQ ID NO:6 and at least one codon of the nucleic acid molecule encoding an amino acid equivalent to at least one of amino acids 681-684 of SEQ ID NO:6 is mutated such the encoded MEKK1 protein is resistant to proteolysis by a caspase after an amino acid equivalent to amino acid 681-684 of SEQ ID NO:6.

63. A method of stimulating apoptosis in a cell comprising introducing into the cell an expression vector encoding a MEKK1 active fragment such that MEKK1 active fragment is produced in the cell and apoptosis is stimulated.

64. A method of inhibiting apoptosis in a cell comprising introducing into the cell an expression vector encoding a protease-resistant MEKK1 protein such that protease-resistant MEKK1 protein is produced in the cell and apoptosis is inhibited.

65. A method of generating an MEKK1 active fragment *in vitro*, comprising: contacting an MEKK1 protein *in vitro* with a caspase protease under proteolysis conditions; and allowing the caspase protease to cleave the MEKK1 protein such that an MEKK1 active fragment is generated.

66. A method of identifying a compound that modulates the apoptotic activity of an MEKK1 active fragment, comprising: providing an indicator cell that comprises a MEKK1 active fragment; contacting the indicator cell with a test compound; and determining the effect of the test compound on the apoptotic activity of the MEKK1 active fragment in the indicator cell to thereby identify a compound that modulates the apoptotic activity of the MEKK1 active fragment.

000050-520000

67. A method of identifying a compound that modulates the proteolytic cleavage of an MEKK1 protein by a caspase protease, comprising:

providing a reaction mixture that comprises an MEKK1 protein and a caspase protease;

5 contacting the reaction mixture with a test compound; and
determining the effect of the test compound on proteolytic cleavage of the
MEKK1 protein by the caspase protease to thereby identify a compound that modulates
the proteolytic cleavage of an MEKK1 protein by a caspase protease.

[illegible]